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(57) Abstract

A pharmaceutical composition for the treatment of cancer, comprising at least one anti-cancer drug selected from mitosene derivatives, anthracyclines, Vinca alkaloids and anthracenediones, in admixture with at least one ganglioside.

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PHARMACEUTICAL COMPOSITIONS FOR THE TREATMENT OF CANCERS

DESCRIPTION

The present invention is concerned with new pharmaceutical compositions for the treatment and amelioration of various types of cancer.

A large number of compounds are already known which can be used with considerable beneficial effect for the treatment cancers. However, almost all compounds at present used for the treatment of cancer are themselves extremely toxic and can give rise to highly undesirable haematological, gastro-intestinal, neurological and other side-effects, especially when administered for long periods of time such as are usually necessary in the treatment of cancers.

Gangliosides are glycosphingolipids which occur in high concentration in the central nervous system. They consist of fatty acids, sphingosine, hexoses, galactosamine and sialic acid. A ganglioside preparation is commercially available under the name

"Cronassial", the composition of which has been found to be as follows:

Monosialotetraesosil ganglioside	(GM- 1)	21%
disialotetraesosil ganglioside	(GD-1a)	40%
disialotetraesosil ganglioside	(GD-1b)	16%
trisialotetraesosil ganglioside	(GT-1b)	19%

Cronassial has been successfully used for the treatment of neurological and associated diseases.

In view of the fact that many of the highly undesirable side effects associated with the therapy of cancers are neurological, we have investigated the possibility of simultaneously carrying out treatments with several different, known compounds which have already been used for the treatment of cancers, together with a treatment with Cronassial.

A number of anti-cancer compounds which we have investigated, include mitosene derivatives, such as mitomycin C and porfiromycin; anthracyclines, such as adriamycin; mitozantrone and vincristine.

In an initial series of experiments, these anti-cancer drugs were tested on the following tumours; sarcoma S180, melanoma B16, leukaemia L 1210 and Lewis lung tumour 3LL. Cronassial was also administered separately. We found that Cronassial had a positive effect on the activity and toxicity of adriamycin, vincristine, mitomycin C and mitozantrone. Because Cronassial reduces the toxicity of the anti-cancer drugs tested, this means that it is possible to administer higher dosages of the drugs to the patients without increasing the risk of toxic side effects.

In a further series of experiments, we prepared mixtures of Cronassial with vincristine, adriamycin, mitomycin C and mitozantrone with the object of simplifying administration.

However, a further series of experiments on the above-mentioned tumours showed that the positive effects achieved when administering Cronassial in admixture with vincristine, adriamycin, mitomycin C and mitozantrone were significantly better than the results achieved by administering Cronassial separately from vincristine, adriamycin, mitomycin C and mitozantrone.

Vincristine is a colourless crystalline solid which dissolves to give a colourless solution, whereas mitozantrone is a blue-black solid which dissolves to give a dark blue solution. When Cronassial is added to a solution of mitozantrone, there is a considerable change of the colour of the solution to a blue/green colour. Further experiments demonstrated that the depth of colour of a solution of mitozantrone is not dependent upon the pH value of solution.

Consequently, it would appear that a chemical reaction or change takes place when Cronassial is added to a solution of mitozantrone and it would appear, therefore, that it is this reaction or change which brings about the significant improvement of the beneficial results which are achieved by the administration of a mixture of Cronassial with mitoxantrone.

Similar experiments with adriamycin showed a colour shift from red to orange/red and with mitomycin C showed a colour shift from violet to blue.

Because vincristine is colourless in solution, no visible changes were observed when Cronassial was added to a solution of vincristine but, since such a mixture also showed a similar significant improvement of the beneficial results, it is reasonable to assume that, here again, a chemical reaction or change takes place.

Although it appears that a chemical reaction or change takes place between the anti-cancer drugs tested and Cronassial, we have not yet been able to ascertain whether some or all of the components of Cronassial participate in this chemical reaction or change. Figures 1 and 2 of the accompanying drawings show the fluorescent spectrum of adriamycin alone and after mixing with Cronassial. The considerable differences between the two spectra provide evidence that a reaction appears to have taken place between the two components.

Therefore, according to the present invention, there is provided a composition for the treatment of cancers, which comprises at least one anti-cancer drug selected from the mitosene derivatives, such as mitomycin C and porfiromycin; the anthracyclines, such as adriamycin;

Vinca alkaloids, such as vincristine; and anthracenediones, such as mitozantrone, in admixture with at least one ganglioside.

The ganglioside used can be, for example, in the form of the above-described "Cronassial".

Since the anti-cancer drugs present in the compositions according to the present invention are administered intravenously in isotonic solution, the compositions according to the present invention are also preferably in the form of an isotonic solution suitable for intravenous administration. In other words, the compositions according to the present invention are prepared and administered in the same way as the anti-cancer drugs present therein.

The weight ratio of anti-cancer drug to ganglioside in the compositions according to the present invention can be about 1:1000, preferably about 1:200 and most preferably about 1:40.

Quite apart from a marked reduction in the toxicity of the known anti-cancer drugs, the compositions avoid variations in the amounts of drug complex administered and also greatly increase the up-take after administration. This means that the compositions according to the present invention result in an amelioration of the undesired side effects but also enable larger doses of anti-cancer drugs to be administered with the danger of increasing the undesired side-effects.

This is clearly shown by Figure 3 of the accompanying drawings, which demonstrates the take up of adriamycin when administered alone and when administered together with Cronassial. The results given in Figure 3 were obtained by treating Chinese hamster ovary cells for one hour and clearly demonstrate that the adriamycin/Cronassial complex behaves differently in solution to adriamycin alone.

The following is a survey of the experiments which have been carried out:

MATERIALS AND METHODS

ANIMALS

Toxicity acute and chronic. Male Swiss Schneider mice with a body weight of 25-30g were used for these experiments. Mice were usually kept in groups of not more than 8 in standard cages. Food and water were supplied ad libidum. The animals were kept in experimental rooms under closely controlled conditions. They were weighed daily. For acute vincristine toxicity in chicks, 48 hour old SPF chicks were used.

Antitumour effects. Male Swiss Schneider mice of 27g body weight were used for the sarcoma S180 experiments: female $C_{57}Bl$ mice of 20g body weight were used for the B16 melanoma experiments; male BDF_1 mice of 30g body weight were used for the L1210 leukaemia experiments; and $C_{57}Bl$ mice of 20g body weight were used for the Lewis lung experiments.

TUMOURS

Sarcoma S180. This tumour has been transplanted in the same strain of mouse for more than 10 years.

Transplantation was done by subcutaneous inoculation of 0.1 ml of a tumour mash made by finely mincing viable tumour tissue and passing it repeatedly through a 26 gauge needle into a sterile Petri dish. 0.1 ml of penicillin (20,000 m/ml) and streptomycin (20,000 m/ml) were added to the mash, as well as neomycin (5 mgs).

Melanoma B16. This tumour was prepared for inoculation in a manner identical to that used for the S180 sarcoma.

L1210. Spleens were removed from animals 7 days after inoculation of L1210 cells and these spleens were finely minced with 1:100 isotonic saline. 0.1 ml of this spleen and leukaemia L1210 cell suspension was then injected subcutaneously into the flank of each BDF_1.

Lewis lung (3LL). This tumour was prepared for inoculation in a manner identical to that used for the S180 sarcoma.

ACTIVE MATERIALS

Vincristine. A standard vial of 'Oncovin' (Eli Lilly Co.) containing 1 mg of vincristine was used. It was made up with the appropriate volume of diluting fluid to give the final concentration required to inject 0.1 ml/10 g body weight.

Cronassial. A solution of this substance, which is a mixture of four gangliosides, was prepared by the addition of a volume of sterile distilled water sufficient to give a concentration of 200 mg/kg in a volume of 0.2 ml. This dose was given to mice of approximately 20g.

Mitozantrone was made up in an appropriate manner. It was given in a volume of 0.2ml when the injections were given intraperitoneally but when given by another route, the volumes given were indicated in the appropriate Table.

RESULTS

Acute toxicity

Large doses of vincristine (i.e. 3.0 mg/kg) given as one intravenous injection to Swiss Schneider mice proved to be fatal for 75% by day 30 (Table 1).

The same treatment but with 200 mg/kg Cronassial given 6 hours before the vincristine resulted in 87.5% deaths by day 30.

It was apparent that Cronassial, under these conditions, gave no protection against the acute toxicity of vincristine.

Acute toxicity experiments using 48 hour old chicks showed that, with high doses of vincristine (6 mg/kg), no acute neurotoxic signs were seen during the first 4 hours in 3/4 chicks but one chick became ataxic and one had preterminal convulsions at 24 hours. All the chicks were dead at 24 hours. In contrast,of those chicks which received 200 mg/kg Cronassial at the same time as the vincristine, only 2/5 had died at 24 hours but only 1 remained alive at 28 hours.

With 4 mg/kg vincristine, 4/5 chicks survived for 24 hours but only 1/5 survived for 28 hours. The chicks which additionally, received 200 mg/kg Cronassial survived better with 3/5 surviving to 28 hours.

With 2 mg/kg vincristine, there was little toxicity and this was not changed when Cronassial was also given.

Therefore, it appears that acute lethal vincristine toxicity can be reduced by Cronassial in large doses. Since even the largest doses of vincristine produced no neurotoxicity, it was not possible to judge whether Cronassial affected this in any way.

Chronic Toxicity

Giving daily injections of vincristine of 0.8 mg/kgfor 5 days was close to the LD_{50}. By giving 200 mg/kg Cronassial at the same time as the vincristine, the number of survivors at 30 days was increased to 100% Almost identical results were obtained by using 1 mg/kg vincristine, even though the drugs were administered for only 4 days. However the total dosage

of vincristine administered in the two experiments (4 mg/kg) was identical. Increasing the cumulative dose of vincristine from 4 to 5 mg/kg increased the mortality of the mice to 100%. With this overwhelming toxicity, there was little or no protection offered by the Cronassial.

The results obtained are summarised in the following Table 1:-

TABLE 1

TOXICITY REDUCTION OF ANTITUMOUR DRUGS - CRONASSIAL & VINCRISTINE

TOXICITY	HOST	DRUG	DOSE mg/kg	DAYS	ROUTE	TIMING	SURVIVORS No.at 30 d	SURVIVAL % at 30 d
Chronic	SN	VCR	0.8	1-5	ip		5/8	62.5
Chronic	SN	(VCR (CRON	0.8 200	1-5 1-5	ip ip	6hrs before VCR	7/7	100
Chronic	SN	VCR	1.0	1-4	ip		3/8	37.5
Chronic	SN	(VCR (CRON	1.0 200	1-4 1-4	ip ip	6hrs before VCR	6/8	75
Chronic	SN	VCR	1.0	1-5	ip		0/6	0
Chronic	SN	(VCR (CRON	1.0 200	1-5 1-5	ip ip	6hrs before VCR	1/5	20
Acute	SN	VCR	3.0	1	iv		2/8	25
Acute	SN	(VCR (CRON	3.0 200	1 1	iv ip	6hrs before VCR	1/8	12.5

CONCLUSION: (1) CRONASSIAL DOES NOT REDUCE ACUTE VINCRISTINE TOXICITY.

(2) CRONASSIAL REDUCES CHRONIC VINCRISTINE TOXICITY.

Antitumour activity with and without Cronassial

Vincristine with and without Cronassial, when tested against a panel of tumours consisting of S180, B16 and L1210, showed that Cronassial did not interfere with any antitumour activity that vincristine might have had as shown by inhibition of tumour growth or increase in mean survival time.

On the contrary, the results obtained against leukaemia L1210 show that the toxic deaths due to vincristine alone can be reduced, when given with Cronassial, and that, therefore, the vincristine effectiveness against L1210 is enhanced with a mean survival time of 10.3 days compared with 7.8 days for vincristine alone. This difference is statistically significant.

The results obtained are summarised in the following Tables 2 and 3:-

TABLE 2

EFFECT OF CRONASSIAL ON VINCRISTINE ANTITUMOUR ACTIVITY

TUMOUR	HOST	DRUG	DOSE mg/kg	DAYS	ROUTE	TIMING	TUMOUR WEIGHTS MEAN (g)
S180	SN	VCR	0.5	1-6	ip		0.64
S180	SN	(VCR (CRON	0.5 200	1-6 1-6	ip ip	6hrs before VCR	0.69
S180	SN	CMC-controls					1.05
B16	C5781	VCR	0.5	1-4,7-11	ip		1.46
B16	C5781	(VCR (CRON	0.5 200	1-4,7-11	ip	6hrs before VCR	1.66
B16	C5781	CMC-controls					2.33
							Mean survival time (days)
L1210	BDF_1	VCR	1.0	1-3	ip		7.8
L1210	BDF_1	(VCR (CRON	1.0 200	1-3 1-3	ip ip	same time as VCR	10.3
L1210	BDF_1	CMC-controls					8.0

CONCLUSION: CRONASSIAL DOES NOT REDUCE VINCRISTINE ACTIVITY

TABLE 3

TOXICITY REDUCTION - CRONASSIAL & VINCRISTINE

TOXICITY	HOST	DRUG	DOSE mg/kg	DAYS	ROUTE	TIMING	SURVIVORS AT 24hrs	28hrs	52hrs
Acute	SPF 48hr chick	VCR	6.0	1	ip		0/4	-	-
Acute	SPF 48hr chick	(VCR (CRON	6.0 200	1	ip	at same time	3/5	1/5	0/5
Acute	SPF 48hr chick	VCR	4.0	1	ip		4/5	1/5	1/5
Acute	SPF 48hr chick	(VCR (CRON	4.0 200	1	ip	at same time	5/5	3/5	1/5
Acute	SPF 48hr chick	VCR	2.0	1	ip		2/2	2/2	1/2
Acute	SPF 48hr chick	(VCR (CRON	2.0 200	1	ip	at same time	2/2	1/2	0/2

CONCLUSION: CRONASSIAL APPEARS TO PROTECT AGAINST ACUTE LETHAL VINCRISTINE TOXICITY IN 48 HOURS OLD CHICKS.

Toxicity reduction of mitozantrone

The following Table 4 clearly shows that it is possible to obtain a toxicity reduction of mitozantrone by Cronassial but that at least 200 mg/kg x 2 are necessary to achieve a definite improvement in survival and the following Table 5 shows the results obtained from a series of experiments to determine the most effective time to give a combination of Cronassial and mitozantone.

Antitumour activity of mitozantrone with and without Cronassial

L1210

From the following Table 5, it can also be seen that there is consistent improvement of the results obtainable with mitozantrone against L1210 when Cronassial is given together with the mitozantrone. This is not a synergistic effect but one which, by reducing the toxicity of mitozantrone, permits high doses of mitozantrone to be given against the L1210.

Lewis lung carcinoma (3LL)

This tumour metastasizes to the lungs when implanted into the flanks of mice. When treated for 5 days between day 5 and 9 after implantation with 3.0 mg/kg mitozantrone, 7/8 mice had died by day 14 so that no assessment could be made of the effect of mitozantrone on the primary or secondary tumours.

On the other hand, a combination of mitozantrone and Cronassial showed that at day 22 the mean primary tumour weight had been reduced from 4.0 g to 1.47 g. and the number of secondaries had been reduced from a mean of 75.1 to 6.1 giving a highly significant T/C of 0.082.

The animals which had been treated with Cronassial only, showed no difference in number of secondaries or in weight of primary tumours from the control animals treated with CMC.

TABLE 4

MINIMUM DOSE OF CRONASSIAL REQUIRED TO REDUCE MITOZANTRONE TOXICITY

TOXICITY	HOST	DRUG	DOSE	DAYS	ROUTE	TIMING	SURVIVORS No. at 30d	SURVIVAL (mean days)
CHRONIC	SN	MTZ	5	1,2	ip		0/6	9
CHRONIC	SN	(MTZ (CRON	5 200	1,2	ip	same time as MTZ	2/6	31
CHRONIC	SN	MTZ	5	1,2	ip		0/6	11
CHRONIC	SN	(MTZ (CRON	5 200	1,2	ip	same time as MTZ	1/6	14
CHRONIC	SN	(MTZ (CRON	5 100	1,2	ip	same time as MTZ	0/6	13
CHRONIC	SN	(MTZ (CRON	5 50	1,2	ip	same time as MTZ	0/6	11

CONCLUSION: 200mg/kg x 2 IS THE MINIMUM DOSE OF CRONASSIAL REQUIRED TO REDUCE MITOZANTRONE TOXICITY

TABLE 5

MOST EFFECTIVE TIME AT WHICH TO GIVE A COMBINATION OF CRONASSIAL AND MITOZANTRONE

TUMOUR	HOST	DRUG	DOSE mg/kg	DAYS	ROUTE	TIMING	SURVIVORS No. at 60 d	SURVIVAL (mean days)
L1210	BDF_1	MTZ	5	0.1.2	ip		1/8	6.14
L1210	BDF_1	(MTZ (CRON	5) 200)	0.1.2.	ip	CR same as MTZ	2/8	14.1
L1210	BDF_1	MTZ	5	1.2.3	ip		2/8	17.1
L1210	BDF_1	(MTZ (CRON	5) 200)	1.2.3	ip	CR same as MTZ	7/8	>60
L1210	DBA	MTZ	5	2.3.4	ip		0/6	16.8
L1210	DBA	(MTZ (CRON	5) 200)	2.3.4	ip	CR same as MTZ	0/6	27.3
L1210	DBA	CMC-controls			ip		0/6	7.2
L1210	BDF_1	MTZ	5	3.4.5	ip		0/8	25.2
L1210	BDF_1	(MTZ (CRON	5) 200)	3.4.5	ip	CR same as MTZ	4/8	42.4
L1210	BDF_1	CMC - controls			ip		0/8	8.7
Cronassial alone as control in all above experiments :							0/8	8.5

CONCLUSION: CRONASSIAL IS EFFECTIVE IN PROTECTING AGAINST MITAZANTRONE TOXICITY.

TABLE 6

CHANGED PROTECTION BY CRONASSIAL OF MITOZANTRONE TOXICITY WITH S180

TOXICITY	HOST	DRUG	DOSE mg/kg	DAYS	ROUTE	TIMING	SURVIVORS No. at 30d	SURVIVAL (mean days)
CHRONIC	SN (+S180)	MTZ	5	3.4.5	ip		0/8	8.8
CHRONIC	SN (+S180)	(MTZ (CRON	5 200	3.4.5	ip	CR same time MTZ		20

CONCLUSION : THE PRESENCE OF S180 DOES NOT INTERFERE WITH THE REDUCTION OF MITOZANTRONE TOXICITY WHICH CRONASSIAL PRODUCES.

Analysis of mechanism of toxicity reduction by Cronassial

All the tests done show that vincristine and mitozantrone toxicity is clearly reduced by Cronassial.

In trying to analyze the mechanism of toxicity protection by Cronassial, the first step is a clear understanding of the toxicity induced by the drugs which have been affected by Cronassial.

The three major systems affected by vincristine are haematological, gastro-intestinal and, in some species, neurological. It is extremely difficult to reproduce the neurotoxicity seen in man in any other species, apart from the cat and chicken, and it is unlikely, therefore, that any reduction of vincristine neurotoxicity by Cronassial would have been seen in our experiments.

However, since there was little effect on acute toxicity in mice but clear evidence on chronic toxicity, it is likely that this could reflect what is seen in man, since it is rare for vincristine to produce acute toxic effects on the CNS or ANS, whereas

the commonly encountered neurotoxicity in the clinic comes from chronic treatment.

On the time scale, therefore, the reduction of vincristine toxicity by Cronassial which we have observed matches that observed in the clinic.

With regard to mitozantrone it would seem unlikely from the results given in the following Tables 7, 8 and 9 that the animals which did not survive had died a haematological death and, therefore, it is unlikely that protection of this system, even if it occurred, played any part in the toxicity reduction of mitozantrone by Cronassial.

TABLE 7

MEAN VALUES OF INDIVIDUAL ANIMALS

RBC x 10^{12}/l

	N	Day 5.	N	Day 7	N	Day 8
CONTROLS	2	7.9		ND	2	7.2
MITOZANTRONE	3	7.3	4	7.0	4	7.1
MOTOZANTRONE & CRONASSIAL	4	7.2	4	7.5	4	6.0

ND = not done

Blood obtained by cardiac puncture - 0.2ml of blood placed into individual sequestrene bottles and blood counts done by Coulter counter.

TABLE 8

MEAN VALUES OF INDIVIDUAL ANIMALS

PLATELETS x $10^9/l$

	N	Day 5	N	Day 7	N	Day 8
CONTROLS	2	632		ND	2	649
MITOZANTRONE	3	631	4	845	4	1058
MITOZANTRONE & CRONASSIAL	4	615	4	873	4	868

ND = not done

Blood obtained by cardiac puncture - 0.2ml of blood placed into individual sequenstrene bottles and blood counts done by Coulter counter.

TABLE 9

MEAN VALUES OF INDIVIDUAL ANIMALS

WBC X $10^9/l$

	N	Day 5	N	Day 7	N	Day 8
CONTROLS	2	9.3		ND	2	5.2
MITOZANTRONE	3	3.5	4	2.2	4	3.2
MITOZANTRONE & CRONASSIAL	4	1.9	4	2.8	4	1.9

ND = not done.

Blood obtained by cardiac puncture - 0.2ml of blood placed into individual sequestrene bottles and blood counts done by Coulter counter

A further series of experiments have also been carried out in order clearly to demonstrate the most effective time for administering mitozantrone and Cronassial and in order to ascertain the toxicity reduction. The results set out in the following Tables 10 and 11 clearly show that there is a remarkable improvement when Cronassial and mitozantrone are mixed together prior to injection in comparison with a separate but simultaneous administration.

TABLE 10

TOXICITY REDUCTION OF MITOZANTRONE DUE TO CRONASSIAL

TUMOUR	HOST	SEX	TREATMENT	DAYS	ROUTE	TIMING	NO. DOSES	DOSE MG/KG	SURVIVORS d.30	SURVIVAL (mean days)
	SN	M	MTZ	1.2.3	iv		x 3	4	0/5	7.8
	SN	M	MTZ + CRON	1.2.3	iv.	mixed as one injection	x 3	4 200	4/5	> 14

Similar experiments have been carried out using a combination of Cronassial with adriamycin and mitomycin C. The results obtained with adriamycin are summarised in the following Tables 11 - 14 and with mitomycin C in Table 15.

In these Tables, MST means median survival time and T/C means treated/control.

TABLE 11

DRUG	DOSE MG/KG	ANIMALS NO/GRP.	DEATHS		SURVIVORS		MST	T/C	COMMENTS
			TOXICITY	L1210	30d	60d			
-	-	8	-	8	-	-	6.8	-	
ADR	4.0	8	-	8	-	-	10.8	159	BDF ♀
ADR + CRON	4.0 200	8	1	6		1	>12.8	>191	20g
ADR	6.0	8	-	8	-	-	9.7	135	
ADR + CRON	6.0 200	8	-	8	-	-	12.1	180	
ADR	8.0	8	-	8	-	-	11.8	174	
ADR + CRON	8.0 200	8	-	8	-	-	13.6	200	

BDF mice inoculated s.c. with L1210. All drugs or control solutions given i.p. in a vol. of 0.2 ml
All drugs given days 1, 2, 3.

TABLE 12

DRUG	DOSE MG/KG	ANIMALS NO/GRP.	DEATHS		SURVIVORS		MST	T/C	COMMENTS
			TOXICITY	L1210	30d	60d			
-	-	8		8	-	-	7.0	-	
ADR	6.0	8	7	1	-	-	9.38	134	BDF ♂ 35g (6/12 old)
ADR + CRON	6.0 200	8	8	-	-	-	10.6	152	
ADR	8.0	8	8	-	-	-	5.8	83	Toxic
ADR + CRON	8.0 200	8	7	1	-	-	8.4	120	
ADR	10.0	8	8	-	-	-	5.5	79	Toxic
ADR + CRON	10.0	8	8	-	-	-	6.0	86	

BDF mice inoculated s.c. with L1210. All drugs or control solutions given i.p. in a vol. of 0.2 ml
All drugs given days 1, 2, 3.

TABLE 13

DRUG	DOSE MG/KG	ANIMALS NO/GRP.	DEATHS		SURVIVORS		MST	T/C	COMMENTS
			TOXICITY	L1210	30d	60d			
-	-	7	-	7	-	-	10.6		
ADR	10.0	7	7	-	-	-	10.3	97	BDF 0 25g (2-3/12 old
ADR + CRON	10.0 200	7	7	-	-	-	16.43	155	
ADR	12.5	7	7	-	-	--	5.4	51	Toxic
ADR + CRON	12.5 200	7	7	-	-	-	5.3	50	
ADR	15.0	7	7	-	-	-	5.1	48	Toxic
ADR + CRON	15.0	7	7	-	-	-	6.0	57	

BDF mice inoculated s.c. with L1210. All drugs or control solutions given i.p. in a vol. of 0.2 ml
All drugs given days 1, 2, 3.

DRUGS : ADR + CRN
TUMOUR: 3LL
PUPRPOSE: Metastases

TABLE 14

DRUG	DOSE mg/kg	ANIMALS No/Grp.	$\bar{p}$ (mean weight)	$\bar{s}$ (mean no)	SURVIVORS 30d	SURVIVORS 60d	MST	T/C	COMMENTS
CONTROL	-	8	5.4g	79.3	-	-	-	-	Experiment terminated d21
ADR	3	8	-	-	-	-	-	-	All ADR alone animals dead by d17
ADR + CRN	3) 200)	8	4.7g	26.7	-	-	-	$\bar{p}$=0.9 $\bar{s}$=0.3	All animals survived to d21

C57Bl mice inoculated s.c. with Lewis lung carcinoma (3LL.) All drugs or control solutions given i.p. in a vol. of 0.2ml.
All drugs given days 6-10

DRUG : Mitomycin C (MMC)
TUMOUR: L1210
PURPOSE: CRN protection v. MMC toxicity

TABLE 15

DRUG	DOSE mg/kg	ANIMALS No./Grp.	DEATHS		SURVIVORS		MST	T/C	COMMENTS
			TOXICITY	L1210	14d	60d			
CONTROL		8	0	8	-	-	7.1		
MMC	5	8	7		1		7.0		
MMC + CRN	5)) 200)	8	3		5		> 14		

BDF mice inoculated s.c. with L1210. All drugs or control solutions given i.p. in a vol. of 0.2ml.

All drugs given days 1, 2, 3.

CLAIMS

1. Pharmaceutical composition for the treatment of cancer, comprising at least one anti-cancer drug selected from mitosene derivatives, anthracyclines, Vinca alkaloids and anthracenediones, in admixture with at least one ganglioside.

2. Pharmaceutical composition according to claim 1, wherein the anti-cancer drug is selected from mitomycin C, porfiromycin, adriamycin, vincristine and mitozantrone.

3. Pharmaceutical composition according to claim 1 or 2, wherein the ganglioside component is monosialotetraesosil ganglioside (GM-1) and/or disialotetraesosil ganglioside (GD-1a) and/or disialotetraesosil ganglioside (GD-1b) and/or trisialotetraesosil ganglioside (GT-1b).

4. Pharmaceutical composition according to any of the preceding claims, wherein the anti-cancer drug and the ganglioside component are present in the form of a complex and/or reaction product.

5. Pharmaceutical composition according to any of the preceding claims, whenever in the form of an isotonic solution for intravenous administration.

6. Pharmaceutical composition according to any of the preceding claims, wherein the weight ratio of anti-cancer drug to ganglioside is about 1:1000, preferably 1:200 and more preferably 1:40.

FIG.1
Adriamycin

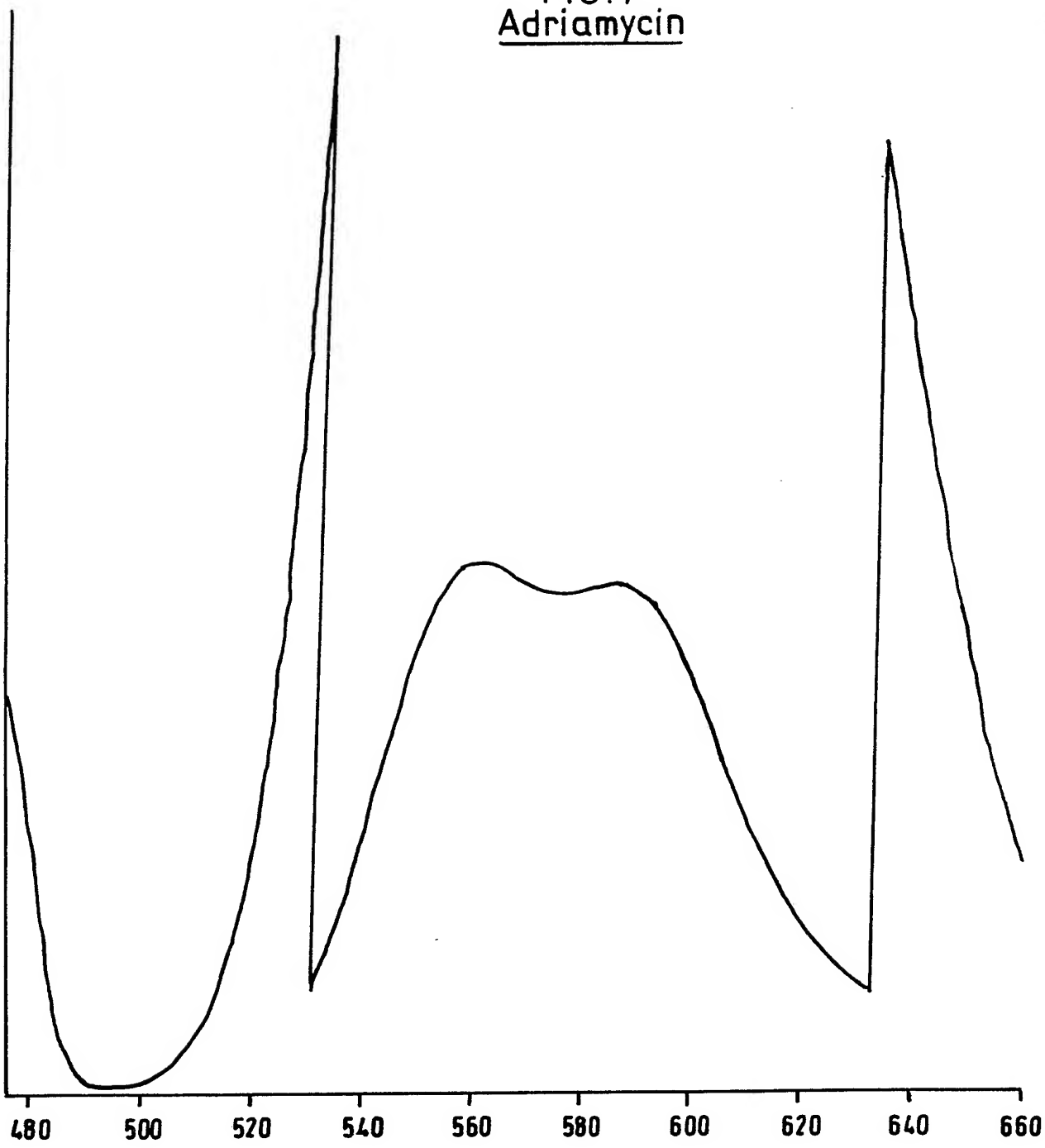


FIG.2

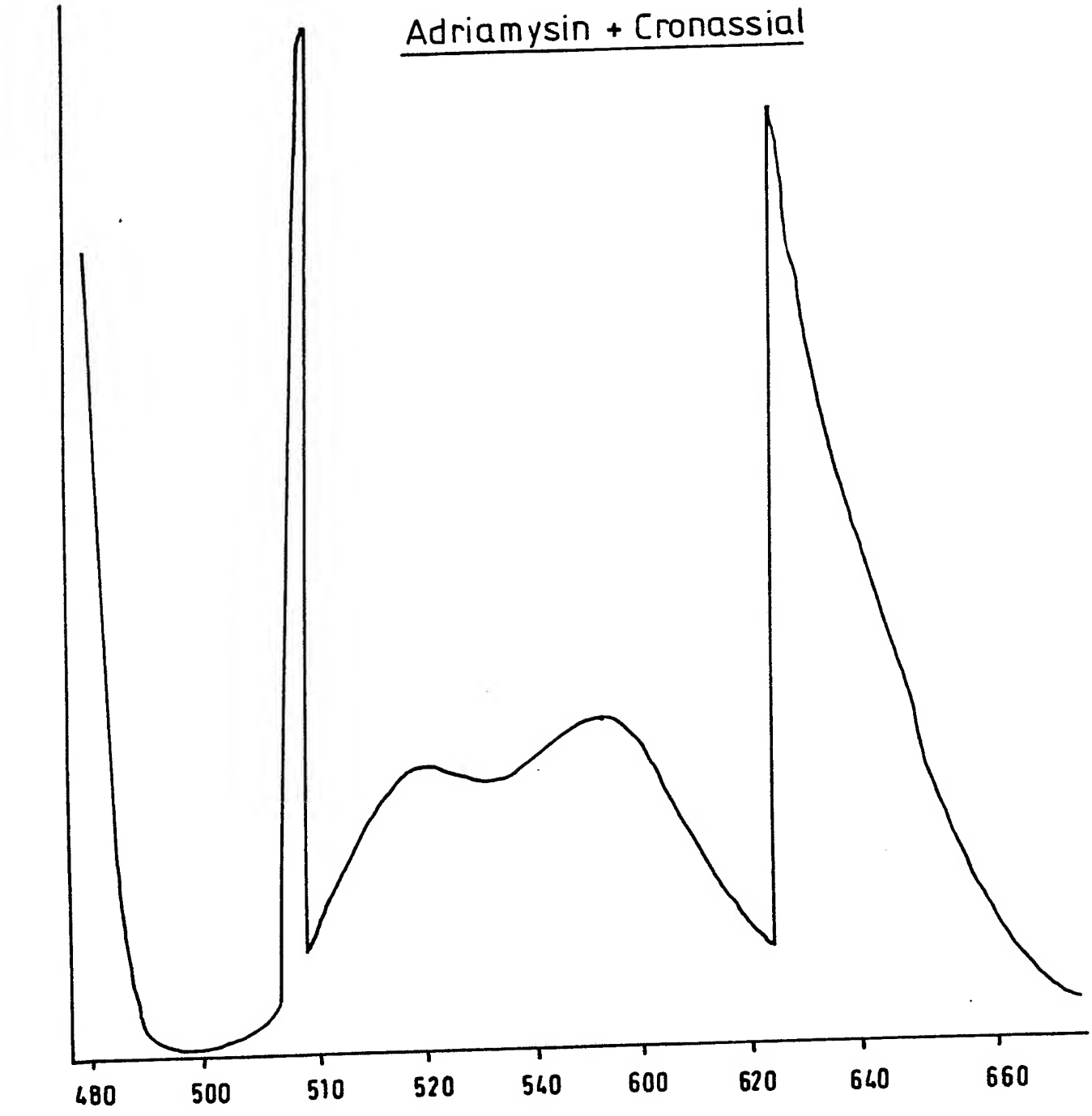
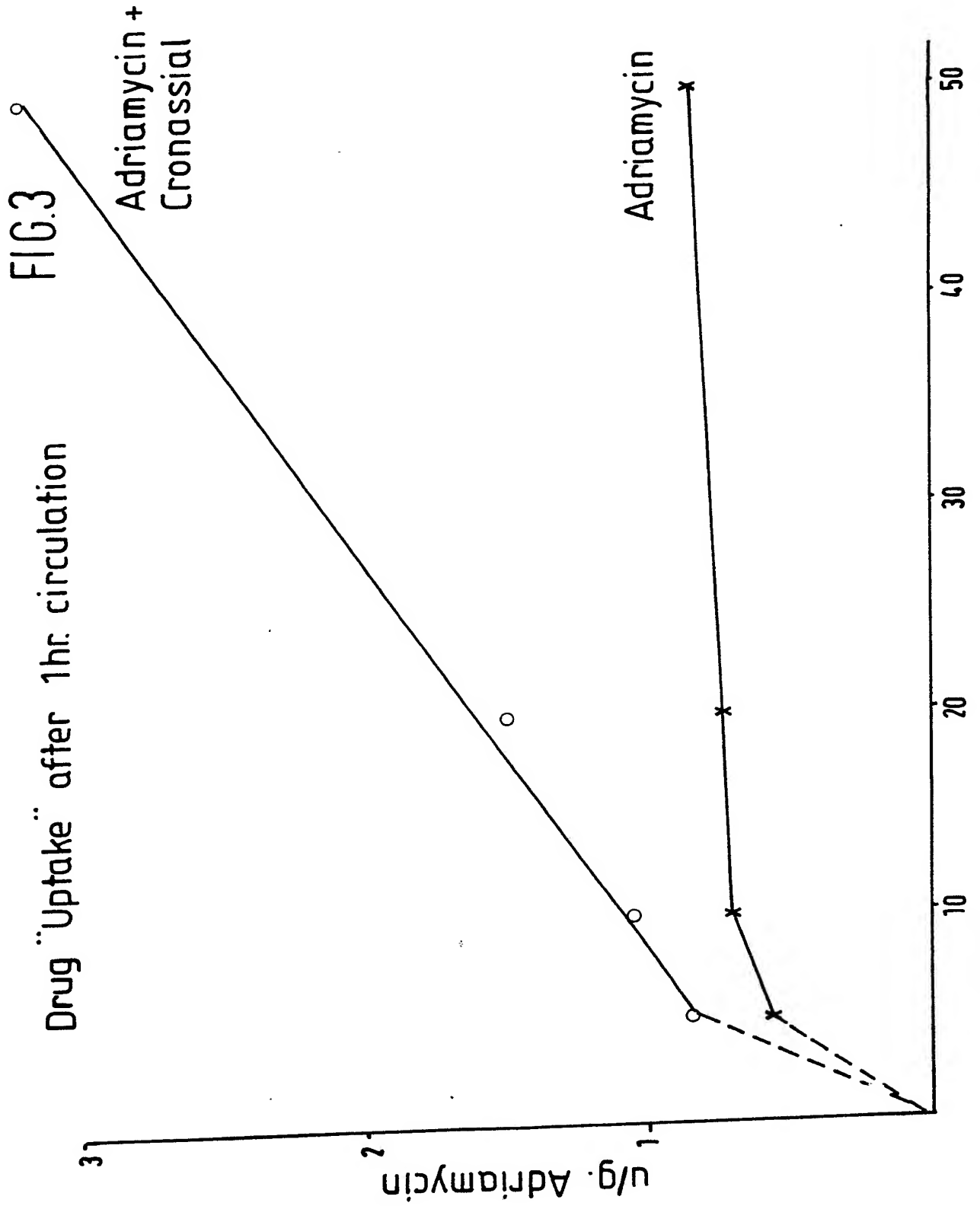


FIG.3


Drug "Uptake" after 1hr. circulation
u/g. Adriamycin
3
2
1
10
20
30
40
50
Adriamycin
Adriamycin +
Cronassial

INTERNATIONAL SEARCH REPORT

International Application No PCT/GB 87/00589

I. CLASSIFICATION OF SUBJECT MATTER (if several classification symbols apply, indicate all) [6]

According to International Patent Classification (IPC) or to both National Classification and IPC

IPC[4]: A 61 K 31/73 // (A 61 K 31/73, 31:475)(A 61 K 31/73, 31:65) (A 61 K 31/73, 31:135)(A 61 K 31/73, 31:40)

II. FIELDS SEARCHED

Minimum Documentation Searched [7]

Classification System	Classification Symbols
IPC[4]	A 61 K

Documentation Searched other than Minimum Documentation to the Extent that such Documents are Included in the Fields Searched [8]

III. DOCUMENTS CONSIDERED TO BE RELEVANT [9]

Category [10]	Citation of Document, [11] with indication, where appropriate, of the relevant passages [12]	Relevant to Claim No. [13]
A	WO, A, 80/01875 (INSTITUT MERIEUX) 18 September 1980 see claims 1,3	1

[10] Special categories of cited documents:

"A" document defining the general state of the art which is not considered to be of particular relevance

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"Y" document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art.

"&" document member of the same patent family

IV. CERTIFICATION

Date of the Actual Completion of the International Search	Date of Mailing of this International Search Report
1st December 1987	14 JAN 1988

International Searching Authority	Signature of Authorized Officer
EUROPEAN PATENT OFFICE	P.C.G. VAN DER PUTTEN

ANNEX TO THE INTERNATIONAL SEARCH REPORT ON INTERNATIONAL PATENT APPLICATION NO. GB 8700589 SA 18336

This annex lists the patent family members relating to the patent documents cited in the above-mentioned international search report. The members are as contained in the European Patent Office EDP file on 22/12/87
The European Patent Office is in no way liable for these particulars which are merely given for the purpose of information.

Patent document cited in search report	Publication date	Patent family member(s)		Publication date
WO-A- 8001875	18-09-80	EP-A,B	0016702	01-10-80
		FR-A,B	2451194	10-10-80
		US-A-	4347244	31-08-82
		DE-A-	2910509	25-09-80